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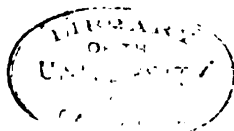


**TOXIC EFFECT OF SALTS ON PLANTS**

by

**Florence N. Magowan**

**A Thesis Submitted in Partial Fulfillment  
of the Requirements for the Degree of Master of Science  
at the  
University of California,  
May, 1907.**







## T O X I C   E F F E C T   O F   S A L T S   O N   P L A N T S .

A few investigations have been made regarding the action of single salts on plants, but in the majority of cases the object has been to determine the limit of endurance or the highest concentration at which certain plants could live in the pure salt solutions. These concentrations varied with the salts used and one could not from the tabulated results tell the difference in growth of plants in the various salts of the same molecular solution, nor would it be safe to assume this difference to vary uniformly in all concentrations. In experiments on *Lumularia*, not yet published, the reverse was found to be true, as the comparative degree of toxicity<sup>ci</sup> of certain solutions at one concentration might be entirely different at another. Again, time was found to be a very important factor in changing this relation, so that experiments lasting only a short period could not be considered conclusive. The work done by previous investigators differs so greatly in method and the results are so scattered that no comparison with my results is possible.

It is the object of this paper to demonstrate by means of curves: (1) the relative degree of toxicity of salts of Ca, K, Na and Mg in equi-molecular solutions; (2) the degree of growth at different concentrations; (3) the variation of results in length of time; (4) the antitoxic effect of salts in each solution; (5) approximately what effect is due to osmotic pressure.

The results obtained here are not claimed to be valid for



all plants, but it is hoped they may be introductory to further experimental work along this line, and throw some light on the whole process of metabolism.

#### MATERIAL USED.

Wheat was taken as being an average well-known agricultural crop, and one with which much experimental work has been done. One variety only was used, the "Early Genesee," which contains a large proportion of starch, and which in preliminary experiments was found to germinate evenly and with a high per cent of germination. Radish and clover seeds were also used for comparison and found to give similar results to wheat.

#### METHOD OF EXPERIMENT.

In practically all experiments that have been made along this line the seeds to be tested were first germinated in distilled water, then placed in the solutions. As the results for this paper were to be the growth made by the seed in the different solutions, this method was not practicable, and the following method was devised whereby the seeds might germinate in the solution from the beginning:

Filter paper equal in length to the circumference of the top of the glass to be used and about 3-1/2 in. in width was folded together once, then folded again, this time having one part about twice the width of the other; so by doubling the smaller part back a trough was made for the seeds. The advantages of this method were: first, seeds germinated in the solution; second, seeds have sufficient air for germination; third, comparison of growth could be easily made:



fourth, seeds and young plants are prevented from sinking; fifth, incrustation of salts on the leaves was prevented, as in preliminary experiments without the filter paper the young plants sank to the bottom of the glass and, wherever the leaves came in contact with the glass, salt collected and quickly killed them.

After the paper strip was folded, and a place cut in one side in order that the level of the solution could be seen and marked, the trough was filled with seeds and the ends of the paper held together with a paper clip. These being all prepared before the solutions were made up, it required only a few moments to remove the clip and place the papers in the glasses, thus avoiding any difference that might arise from not starting all at the same time.

Twenty or more seeds were placed in each solution, and the average ones were measured to ascertain the total length of roots in millimeters per grain, and the result given here is the average of those measured. Special care was taken in selecting the average ones, but in general the germination was quite uniform.

Duplicate experiments were repeated several times, and, while one series may have made a better growth than another, the comparative order of toxicity was not changed, and in all concentrations final measurements were made when the roots had reached the limit of growth.

To aid the rootlets of the radish and clover to penetrate the filter paper, a tracing wheel was run along the bottom of the "trough." While this was not necessary, it was found beneficial to



the wheat also, making it easier to remove the plants from the paper, as well as for roots to make their way through.

After the first series it was thought worth while to determine the death rate of the salts in question. With this object in view, five to ten average plants in each concentration were after six or seven days fastened to spring clothes-pins by means of rubber bands, and the pins were placed on the glasses with the radicles immersed, but without allowing the clothes-pins to touch the solutions. If, upon transferring a plant to a glass of distilled water, it was found to make no growth, it was considered dead.

The solutions were titrated against standard silver nitrate solution in the beginning to avoid any error, and great care was taken in making the various concentrations.

To keep the solutions at the same strength was a problem which received careful consideration. As little salt would be taken by the plant the first few days, it was found perfectly safe to replenish the loss by addition of distilled water. When, however, by titration the solutions were found too weak, they were replaced by fresh solutions.

#### SALTS USED.

Several concentrations of Chlorides of Ca, K, Na and Mg were used, these salts being taken because they represent the bases most common in the soil.

Control experiments with tap and distilled water were always carried on with each series. The plants made an excellent





growth in distilled water, which of course was all important in this work, in order that the toxic or stimulating effect of the salts could be determined. The water was distilled with especial care to avoid all traces of metals in solution.

Van't Hoff's solution, which is assumed to be non-toxic, formed part of every series, being of the same concentration as the salts, to demonstrate how much of the action was due to osmotic pressure. In these experiments only one anion, chlorine, was used, so that the effect would be due solely to the cations.

The curves represent the total growth of root hairs in millimeters per wheat grain in the concentration given. As seen by Fig. I., little growth took place at .18 M in Ca, Na and K Chlorides and none whatever in  $MgCl_2$ , where the small growth throughout the series<sup>^</sup> of concentrations is undoubtedly due, in part, to the fact that the molecule of  $MgCl_2$  when dissolved dissociates into three ions instead of two, as in the case of NaCl and KCl; hence,  $MgCl_2$  solutions have a higher osmotic pressure than equi-molecular solutions of NaCl and KCl, and consequently prevent plants from taking up so much water.

All the more striking is the behavior of  $CaCl_2$ , which in spite of high osmotic pressure (approximately as high as  $MgCl_2$ ) nevertheless permits greater growth than NaCl or KCl; hence it must be much less toxic than any of the other salts, while  $MgCl_2$  is the most toxic.

Another striking fact brought out in Figure I. is that the salts, each of which is poisonous by itself, when combined together in van't Hoff's solution form a mixture in which the



plants make a splendid growth. This is not the phenomenon familiar to us in water cultures and in soil fertilization, where the addition of necessary salts gives a greatly increased growth, for at this stage of development no salts whatever are necessary and plants make a better growth in distilled water than in any of the solutions shown in the curve.

The toxic effect of the salts was noticeable principally in the radicles, as the tops were comparatively the same in the different salts, with the exception of those in the stronger concentrations of Mg, in which little growth of either radicles or tops occurred. The order of germination was also noteworthy. The seeds in NaCl and KCl were the first to send out roots and remained in advance of those in  $\text{CaCl}_2$  for several days.

Loeb found the action of K, Na and Ca salts upon the absorption of water by the muscle to be analogous to the behavior of the various soaps; that is, the presence of Ca retarded while K favored the absorption of water. Such was found to be true with wheat grains, equal weights of which were placed in equi-molecular solutions of Ca, K, Na and  $\text{MgCl}_2$ . After periods of twenty-four, forty-eight and ninety-six hours they were removed, dried between filter paper and weighed. Those in KCl solutions absorbed water much more rapidly than in  $\text{CaCl}_2$  and  $\text{MgCl}_2$ , while NaCl occupied a midway position. This order continued true for the first forty-eight hours; then the seeds in all the solutions weighed approximately the same, indicating the cause of slower growth might be due to rate of absorption.

For the first four or five days the wheat in the  $\text{MgCl}_2$  so-



lutions made a growth almost equal to those in  $\text{CaCl}_2$ , but after fifteen days the radicles in  $\text{MgCl}_2$  series had reached their limit of growth, while those in  $\text{CaCl}_2$  .08 M were still living and <sup>even</sup> in .12 M new roots were produced when transferred to distilled water.

It is significant to note the effect of these salts on the general appearance of the roots:  $\text{CaCl}_2$  favored the development of root-hairs, a fine growth beginning in the .06 M solution and continuing throughout the lower dilutions of the series; also many side branches are produced, making a thrifty-looking root-system.

At no concentration of KCl and NaCl were the root-hairs developed equally to  $\text{CaCl}_2$  .06 M, and those in the weaker solutions were decidedly unthrifty in appearance. KCl gave the roots a dull white color, but as to branching was next to  $\text{CaCl}_2$  although the branches were thicker, shorter and most abundant in .02, .04 and .06 M concentrations, while in  $\text{CaCl}_2$  branching was practically the same throughout. NaCl developed a few stunted-looking branches, and after twenty days the roots were of a yellow color in all the higher concentrations down to .02 M.  $\text{MgCl}_2$  produced no branches nor root-hairs in concentrations .08 M to .005 M, but in .001 M root-hairs were developed, and in .0001 M branches were sent out similar to those in  $\text{CaCl}_2$  solutions.

Figures II., III. and IV. show a combination of two salts far may be better than either salt alone. This is true of  $\text{KCl} + \text{CaCl}_2$  and of  $\text{NaCl} + \text{CaCl}_2$  and also of  $\text{MgCl}_2 + \text{CaCl}_2$  except in the higher concentrations.

These results agree with those obtained by Professor Oster-



hout in his experiments with marine and fresh-water algae, where the salts just mentioned produced, alone, little or no growth, but in the combinations just given ( $\text{KCl} + \text{CaCl}_2$ ,  $\text{NaCl} + \text{CaCl}_2$ ,  $\text{MgCl}_2 + \text{CaCl}_2$ ) they made good growth. In other words, an antagonistic effect was shown. But when Ca was omitted and two or even all three of the other salts were combined, there was no advantage over the single salts. Since my experiments, as far as they were carried, agreed with these results, the presumption is that for the wheat, as for the algae, the presence of Ca in a combination is helpful in all instances, while the omission of Mg or Na or K would be scarcely noticeable. This importance of Ca is in accord with Professor Hilgard's statement that "lime is a governing factor" and "a lime country is a rich country," as *lime* will counteract the toxic effect of an excess of the other salts present.

Figure V. shows that the addition of Ca greatly diminishes the toxicity of each single salt but does not change the relative toxicity of the salts when compared with one another. Taking 100  $\text{KCl}$  + 20  $\text{CaCl}_2$  as a non-toxic combination exerting an influence chiefly through its osmotic pressure, it would be possible by this curve to estimate what per cent of the results here stated are due to osmotic pressure and what per cent due to toxicity.\*

The distance from the line AB, representing distilled water, to the curve of  $\text{KCl} + \text{CaCl}_2$  would be the per cent due to osmotic pressure, while the distance of the other curves from  $\text{KCl} + \text{CaCl}_2$  would be the per cent due to toxicity. For example, 100  $\text{MgCl}_2$  + 20  $\text{CaCl}_2$  at .12 M

\* This is of course only -8- true when the osmotic pressure of the solutions are nearly the same or when both





produces an effect of which 64.5 % is due to osmotic pressure and 35.5 % due to toxicity. The same can be done with Figure I., taking the van't Hoff's solution curve as representing the effects of osmotic pressure and measuring to the distilled water mark. It is very noticeable that in lower concentrations the effect of osmotic pressure decreases as the toxicity increases.

### RESULTS.

1. Each of the principal soil bases (Na, K, Mg, Ca) is toxic when it alone is present. The following is the order of toxicity of their chlorides: first,  $MgCl_2$ ; second, NaCl: third, KCl, and fourth,  $CaCl_2$ .

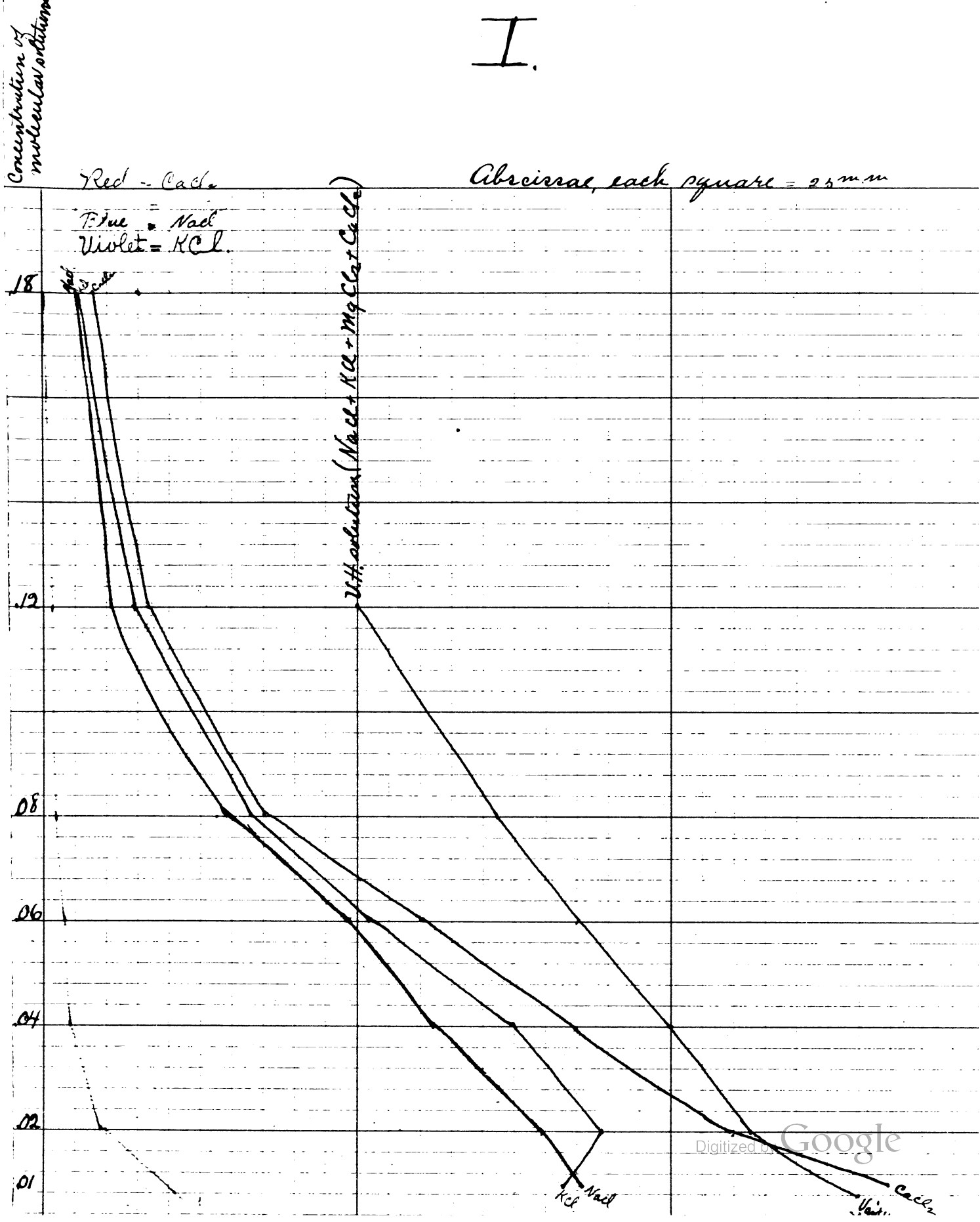
2. The deleterious influence of Na, K, and Mg when alone or in combinations can be counteracted by Ca.

3. Osmotic pressure is the most important factor in suppressing growth in the strong solutions, but the toxic action is the most important factor in the weaker solutions.

4. Experiments lasting but a short time (such as have been made by all previous investigators of toxic action of salts) can not be considered conclusive, for after six days plants in NaCl and KCl made more growth than in  $CaCl_2$ , but after twenty-five or thirty days the plants in  $CaCl_2$  were in advance of all the others.



# I.



1"

# II

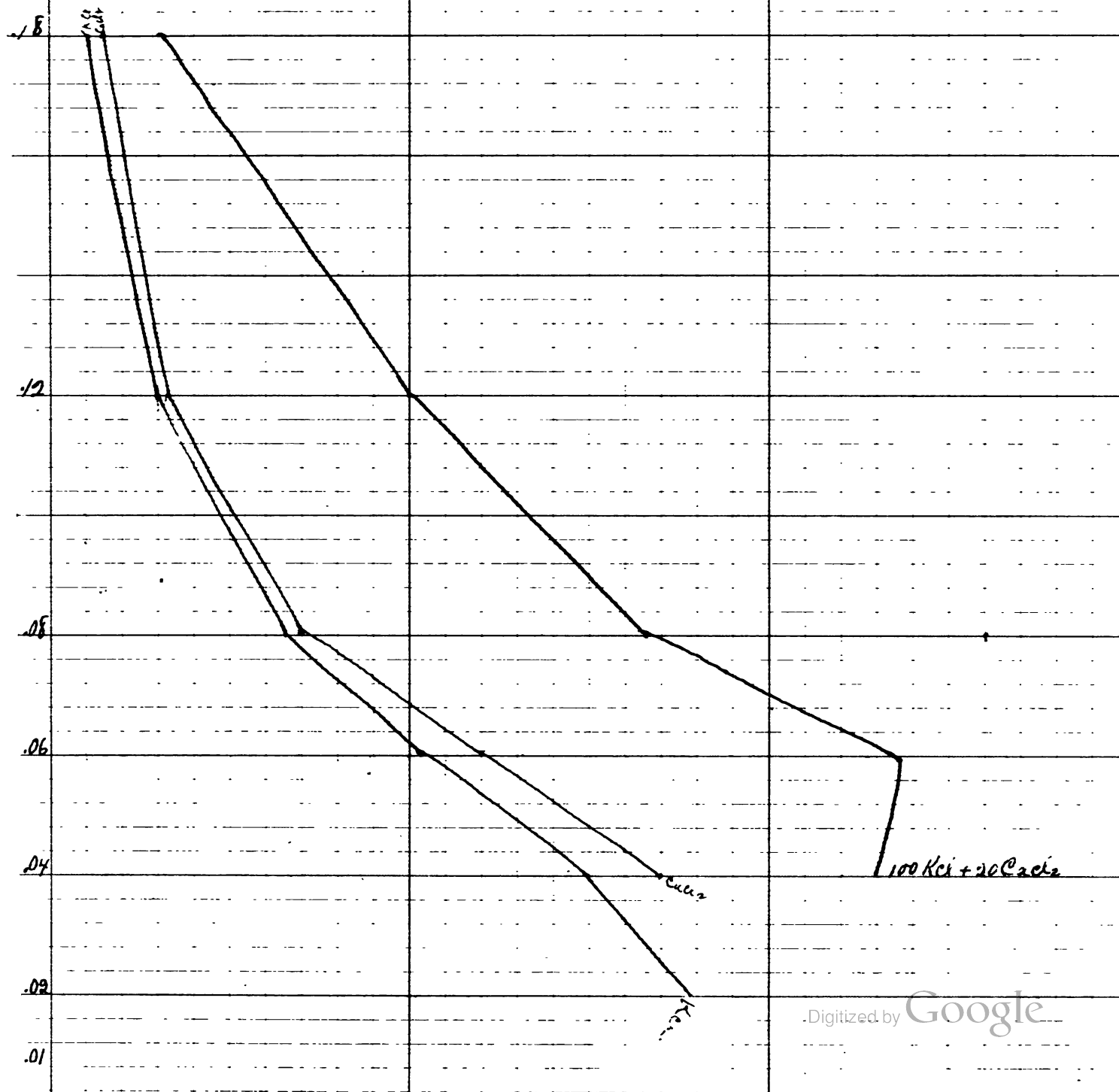
Concentration of  
molecules at Sol.

Abscissa - each square = 25 m. m.

Black =  $KCl + CaCl_2$

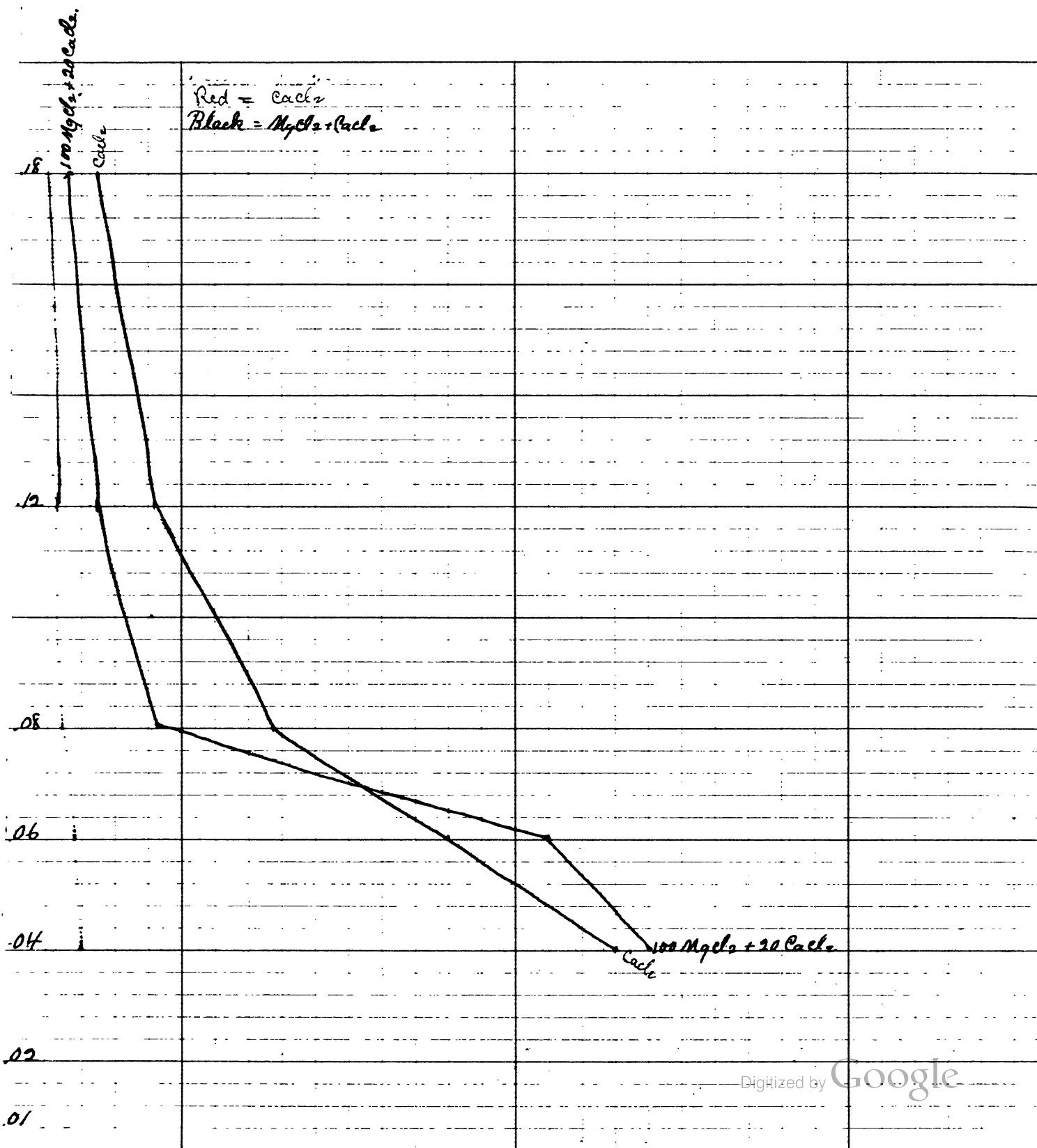
Red  $CaCl_2$

Violet =  $KCl$





IV

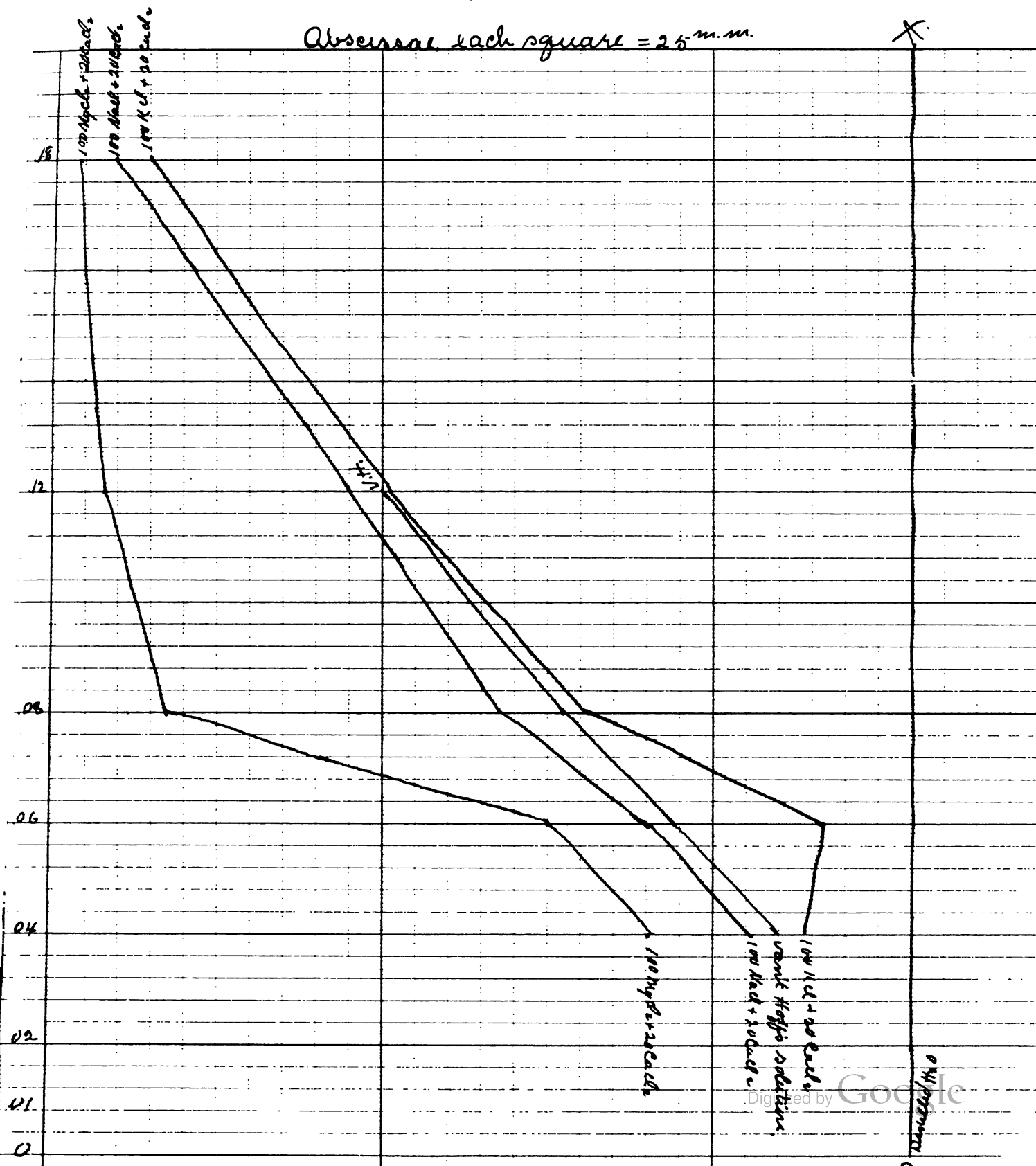


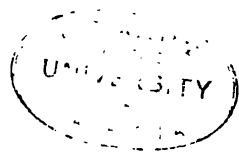




V.

Abscissae each square = 25 m.m.

















































































































































































































































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